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The goal of this research training program was to produce highly qualified scientists for careers as independent investigators in the field of breast cancer. During the last 20 years, there has been a fundamental revolution in the understanding of molecular and cell biological concepts related to cell growth, function and tumorigenesis. To utilize what has been learned and to continue future progress in the area of breast cancer requires the continued availability of well-trained, innovative and committed scientists. This program represents an interdepartmental training program involving thirteen investigators from six departments. Trainees were predoctoral fellows with backgrounds in biochemistry, cell and molecular biology, molecular genetics and molecular virology. The training program provided trainees with additional foundation in carcinogenesis and breast cancer. In addition to the core curriculum taken by the predoctoral fellows in their respective academic departments, program enhancement was provided through trainees' participation in a graduate course on "Molecular Carcinogenesis", a Breast Cancer Seminar Series, and participation at national meetings and local seminars. Fourteen predoctoral trainees were enrolled over the past four years with six predoctoral trainees currently enrolled in the training program. The training program was considered a success with nine publications and seven meeting abstracts resulting so far with additional manuscripts in preparation.

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Daniel Medina June 25, 1998
PI - Signature Date

4. TABLE OF CONTENTS

5. INTRODUCTION

Breast cancer is a complex disease whose ultimate understanding will require the integration of facts resulting from a multidisciplinary approach. Continued basic science research will provide a fuller understanding of the basic mechanisms of breast cancer which is necessary to conquer the disease in humans. In order to have the scientific human armamentarium to further this understanding, this training grant focused on producing qualified scientists for careers as independent investigators in the area of breast cancer. The rationale for a targeted training grant in breast cancer is based on the belief that the elucidation of how oncogenes, tumor suppressor genes, hormones and growth factors act at the molecular level and as developmental-specific agents are critical questions directly relevant to the etiology, prevention, diagnosis, treatment and prognosis of human breast cancer. The training program has drawn together individuals who have an established research and training background in the mammary gland with individuals who have a research and training background in cell biology, molecular endocrinology, molecular biology, molecular virology, viral oncology, molecular genetics and biochemistry. The strength of the program was two-fold. First, the program gathered together members of diverse disciplines to focus on the training of predoctoral students for careers in an area which, by its biological nature, is multi-disciplinary. Second, the program introduced new intellectual approaches and insights to the problem of breast cancer which will be continued by the next generation of research scientists.

The design of the training program provided for trainees to be exposed to clinical problems and recent advances as well as the multi-disciplinary approaches to answering fundamental questions related to breast cancer research. The familiarity and close proximity of the training faculty facilitated and encouraged the development of a new generation of research scientists who will be able to understand the problem of breast cancer at a more complex level and from a multi-disciplinary orientation.

6. BODY OF PROPOSAL

A. Trainees

The goal of this training program was to provide an environment for training in breast cancer research. To foster this goal, candidate graduate students had to meet a minimum set of requirements. Graduate students had to be at least in their second year of graduate school and had selected a thesis problem focusing on an aspect of mammary gland growth, differentiation and/or cancer. These students would be supported for two years by the training program provided they maintained satisfactory progress in their research program and they participated in the biweekly journal clubs and attended the course "Molecular Carcinogenesis". The progress and status of the first six students, and an additional student who just graduated, is summarized in Table 1. The summary of their research progress was provided in the previous progress reports, therefore the status only of these students is listed in Table 1. Four of the students have graduated or will graduate by August 1998 with the two others scheduled in 1999. Deana Roy is a special status graduate student. Deana married a fellow student in 1997 and moved with him to Montreal, Canada. Currently, she is continuing her graduate studies under joint sponsorship of McGill University and Baylor College of Medicine and will obtain her Ph.D. with Dr. J. Rosen as mentor. Each student satisfactorily completed the requirements of the training program in the two years of support and maintained an active research involvement in breast cancer-related problems.

In July 1996, a second set of students rotated on the training program. Funding considerations resulted in one student, Jeff Jones, being maintained on the training grant for a second period of two years. A second student, Lilia Stepanova, participated fully in the program for 1996-1997, and then was awarded an individual predoctoral fellowship from the DOD breast cancer program. A third student left graduate school in June 1997, and his position was filled by another third year student from the same laboratory. The current six students, their departmental affiliation, major advisor, thesis problem and an Abstract of their research is provided below:

a. Jeffrey M. Jones, Department of Molecular Virology, Lawrence A. Donehower, Ph.D., "The role of p53 and p21 in mammary tumorigenesis and growth in the *Wnt*-1 transgenic mouse.

ABSTRACT

In order to specifically address the role of loss of the *p53* gene in breast cancer and how this loss affects tumor progression we have developed a mammary tumor specific mouse model. We have crossed our *p53* -deficient mice, which are highly susceptible to a wide variety of early onset spontaneous tumors, to *Wnt-1* transgenic mice. The *Wnt-1* transgenic mice carry a MMTV LTR driving mammary specific expression of a *Wnt-1* transgene. The ectopic expression of this growth factor results in the development of mammary tumors in these mice. The *Wnt-1* transgenic mice are a well established mammary tumorigenesis model. By crossing these two strains of mice we have established a new model in which we can specifically address the role of *p53* in mammary tumorigenesis.

Using this model we have established that tumors which are deficient for *p53* arise earlier and grow more rapidly than tumors with one or two wildtype alleles. The question we then wanted to ask is which function of *p53* is responsible for this growth inhibitory function. Using immunohistochemical techniques (TUNEL) we have shown that there is no difference in the levels of apoptosis observed in these tumors based on the presence or absence of *p53*. We have observed a significantly larger fraction of cells proliferating in *p53*-deficient tumors than in *p53*-positive tumors using both flow cytommetry and mitotic figure counts. This suggests that *p53s* mechanism of inhibiting tumor growth rate in this model system is through its capacity to induce a cell cycle arrest and not through its ability to induce apoptosis.

Based on the observation that *p53's* primary mechanism of inhibiting tumor growth rate in this system is through its capacity to inhibit cell cycle progression we have crossed a second set of mice to test this observation in another biological system. We have crossed the *Wnt-1* transgenic mice to the *p21*-deficient mice. *p21* is a cyclin dependent kinase inhibitor which is activated by *p53* in order to cause a G1 cell cycle arrest. By breeding *Wnt-1* transgenic / *p21*-deficient animals we hope to show that the lack of *p21* is enough to accelerate tumor growth rate even in the presence of wildtype *p53*.

We have shown in this system that the presence or absence of *p21* in the *Wnt-1* transgenic mouse system has no effect on when tumors arise. However, tumors grow significantly faster in the *p21* +/- animals than in animals with two wildtype or two null alleles for this gene. We attribute the lack of accelerated tumor growth in the *p21* -/- animals to the necessity of *p21* to act as a adapter molecule between the cyclin and the CDK. Without *p21* there is reduced formation of active cyclin/CDK complexes. The accelerated tumor growth observed in the *p21* +/- animals is attributed to the reduced *p21* complement not allowing cells to produce sufficient quantities of *p21* to inhibit cyclin/CDK action. We have shown increased cyclin/CDK activity in tumors from *p21* +/- animals versus *p21* +/+ and -/- animals by Rb phosphorylation assay.

b. Michael Mixon, Department of Cell Biology, Daniel Medina, Ph.D., "The role of Brca1 in mouse mammary cancer".

ABSTRACT

While the function of *Brca1*, a probable tumor suppressor involved in familial breast and ovarian cancer, has yet to be determined, study of this gene in the mouse system may provide insights relevant to the biological function of this protein and its involvement in human disease. Because germline mutation of this gene in the human strongly predisposes to breast cancer, analysis of expression of the endogenous *Brca1* transcript has been performed on the mouse mammary gland at various stages in gland development. Expression is easily detectable in the virgin gland, increases approximately twofold by midpregnancy, and then declines to levels not significantly different than those in the virgin gland after completion of involution after weaning. While contrary to published observations of a persistent increase in expression after involution, this expression pattern has been observed repeatedly with probes to several different regions of the gene. This observed pattern is not a reflection of genetic background, as the same trends are seen in both the Balb/c mice we use and the FVB mice of the conflicting report. While normal *Brca1* expression is high in rapidly proliferating cells that are also undergoing subsequent differentiation, the possibility existed that *Brca1* expression may be

altered during the progression from normal gland to tumor. To see whether this was the case, expression of *Brca1* was checked in nine mammary hyperplasias and paired tumors derived from these outgrowths. Although expression of *Brca1* mRNA is reported to decrease upon progression to invasive breast cancer in humans, this pattern was not observed in the mouse. *Brca1* levels in the tumors were at least as high as in the hyperplasias, and possibly higher. As progression to tumorigenicity arguably reflects a loss of differentiation, this indicates that *Brca1* expression can be decoupled from the differentiated phenotype.

While several splice variants of *Brca1* lacking most or all of the large exon 11 have been detected in the human, a single splice variant lacking almost the entire exon 11 was isolated from an immortal but nontransformed MME cell line. Expression of this variant parallels full-length in all the aforementioned cases. To study the effects of exogenous expression of *Brca1*, the complete sequence was constructed from overlapping genomic cDNA clones and introduced into a mammary tumor-derived cell line. Stable cell lines were generated and tested for tumor-forming ability in the cleared fat pad pending characterization of transgene expression. Of seven cell lines tested, one has in two separate experiments formed tumors more slowly than vector-only controls. This cell line has been singled out for characterization and is being analyzed for extent of *Brca1* overexpression and copy number of transgene.

One possible role for *Brca1* may be suggested based upon its association with homologs of the Rad52 epistasis group proteins. Mutations in these genes frequently confer radiosensitivity upon cells; overexpression of wild-type protein in some cases can provide radioresistance. If *Brca1* is involved in processing of DNA damage, overexpression of *Brca1* may lead to an improved DNA damage repair response. This will be tested for radiation-induced double-stranded break repair in the cell line expressing the most exogenous *Brca1*.

Expression analysis is incomplete unless looked at at the protein level. Repeated analysis of available Ab's has shown that while recognizing IVT *Brca1* and possibly wild-type *Brca1* in cell extracts, these Ab's are not sensitive enough for tissue analysis nor specific enough for IHC. For this reason, a polyclonal antibody to a *Brca1* peptide has been commercially generated and is currently being tested for sensitivity and specificity.

c. Shannon Lindsey Wyszomierski, Department of Cell Biology, Jeffrey M. Rosen, Ph.D., "Role of STAT5 in mammary epithelial cell signaling".

ABSTRACT

The ß-casein promoter contains a composite response element (CoRE) which is sufficient to confer appropriate hormonal induction in mammary epithelial cells of an attached gene in response to insulin, hydrocortisone (HC) and prolactin (PRL). Because the transcription factor binding sites in this CoRE are well characterized, it provides an ideal model for analyzing protein-protein interactions between transcription factors. STAT5, C/EBP, and glucocorticoid receptor (GR) are all needed for strong activation of the β-casein promoter during lactation. The LIP isoform of C/EBPβ (which acts in a dominant negative fashion) and YY1 are believed to play a role in repression of the β-casein promoter in the virgin, during most of pregnancy and during involution of the mammary gland. STAT5a2, a naturally occurring splice variant of STAT5a, may also play a role in repression of β-casein . It has been found to act as a

dominant negative to STAT5 in some situations. Similar collections of transcription factor binding sites are found in other promoters. Understanding how these transcription factors interact and which interactions are occurring in the normal mammary gland is, therefore, important in order to understand how they may be altered during mammary carcinogenesis.

My work has been focused on the GR and STAT5 interaction. Previous studies, in other laboratories, have demonstrated that STAT5 directly interacts with GR both in transfected COS cells and in HC11 mammary epithelial cells and that GR acts synergistically with STAT5 to enhance transcription from the ß-casein promoter. I have performed extensive electrophoretic mobility shift analysis using an oligonucleotide containing the STAT5 binding site in the ß-casein CoRE with extracts from cells transfected with the prolactin receptor, STAT5 and GR. I have discovered that GR enhances STAT5a binding to GAS site following prolactin and hydrocortisone treatment. The level of STAT5a protein in the cells remains constant. The GR- enhanced STAT5a- containing complex is more resistant to oligonucleotide competition. Additionally, GR allows STAT5a to stay phosphorylated and capable of binding DNA for a longer period of time after PRL treatment. We believe the protein-protein interaction between GR and STAT5 decreases the interaction between STAT5 and an unidentified tyrosine phosphatase believed to be responsible for STAT5 inactivation. I have shown that this is not a general effect of steroid receptor/ STAT5 interactions by demonstrating that co-transfection of the estrogen receptor leads to an overall decrease in STAT5a tyrosine phosphorylation without effecting the dephosphorylation rate. The proposed mechanism for GR/ STAT5 transcriptional synergy is through cooperative effects of the transactivation domains of the two proteins. My work adds a role for the enhancement or stabilization of the DNA bound activation complex in the transcriptional synergy exhibited by STAT5 and GR. Additionally, it provides an interesting example of how proteins acting together at a CoRE may modulate each other's activation state.

I have recently initiated work to better understand the protein-protein interactions involved in repression of the β-casein gene. I have demonstrated that the LIP isoform of C/EBPβ is a very potent inhibitor of the β-casein promoter. The portion of C/EBPβ which is retained in LIP is known to interact with YY1 and YY1 is known to interact with all of the human histone deacetylases identified to date. Because of the proximity of a YY1 site and a C/EBP binding site in a region of the β-casein promoter implicated in repression, we hypothesize that YY1 and LIP form a complex which recruits a histone deacetylase to the promoter and keeps it inactive. Experiments to test this hypothesis are in progress. The role of STAT5a2 is also of interest. Previous studies on carboxy- truncated STAT3 have shown that it can act either as dominant negative transcription factors or as positive transactivators depending on the promoter and co-factors present. In COS cells, STAT5a2 is capable of transactivation only when GR is present. When GR is not present, STAT5a2 inhibit STAT5 activation of the β-casein promoter . My work has demonstrated that this effect is cell type specific. In CHOk1 cells, which contain endogenous GR and STAT5a, STAT5a2 is still an effective inhibitor of the β-casein promoter. Preliminary results indicate that this inhibition cannot be reversed by adding exogenous GR to the cells. STAT5a2 appears to be a less potent inhibitor of the β-casein promoter than LIP and comparison of the mechanisms of these two inhibitory proteins should be interesting.

d. Xiaohong Leng, Department of Biochemistry, J. Wade Harper, Ph.D., "Role of Cdk-induced phosphorylation in cell cycle control".

ABSTRACT

The development of cancer is the result of uncontrolled cell proliferation. In mammalian cells, the retinoblastoma protein (Rb) is thought to negatively regulate progression through the G1 phase of the cell cycle by its association with the transcription factor E2F. Rb-E2F complexes suppress transcription of genes required for DNA synthesis, and the prevailing view is that phosphorylation of Rb by complexes of cyclin-dependent kinases (Cdks) and their regulatory cyclin subunits, and the subsequent release of active E2F, is required for S-phase entry. This view is based, in part, on the fact that ectopic expression of cyclin-Cdks leads to Rb phosphorylation and that this modification correlates with S-phase entry. In Drosophila, however, cyclin E expression can bypass a requirement for E2F, suggesting that cyclins may activate replication independently of the Rb/E2F pathway. We thought to examine whether Rb phosphorylation is a prerequisite for S-phase entry in Rb-deficient SAOS-2 osteosarcoma cells, using a commonly used cotransfection assay. We find that a G1 arrest in SAOS-2 cells mediated by an Rb mutant lacking all 14 consensus Cdk phosphorylation sites is bypassed by coexpression G1-specific E-type or D-type cyclin-Cdk complexes, and that injection of purified cyclin-Cdks during G1 accelerates S-phase entry. Our results indicate that Rb phosphorylation is not essential for S-phase entry when G1 cyclin -Cdks are overexpressed, and that other substrates of these kinases can be rate-limiting for the G1 to S-phase transition. These data also reveal that the SAOS-2 cotransfection assay is complicated by Rb-independent effects of the coexpressed Cdks. Besides, biochemical analysis of p107, another member in Rb pocket protein family, has been carried out. CyclinD1/Cdk4 shows specific phosphorylation pattern of p107 compared to that of CyA/Cdk2 and CyclinE/Cdk2 *in vitro*. The biological significance of site-specific phosphorylation by various G1 Cdks in the term of controlling cell proliferation is being studied.

e. Hong Wang, Department of Biochemistry, Stephen J. Elledge, Ph.D., "Role of Sdt1 in S-phase checkpoint signalling".

ABSTRACT

Cell cycle checkpoints are regulatory pathways that control the order and timing of cell cycle transitions. In *S. cerevisiae*, in response to a DNA replication block, the S-phase checkpoint pathway delays the onset of mitosis and induces the transcription of genes whose products facilitate DNA replication, therefore, avoiding chromosome breakage and chromosome loss. Mutations in genes required for the S-phase checkpoint display premature anaphase before DNA synthesis is completed. Among these genes, *POL2*, encoding the catalytic subunit of DNA polymerase II(e), may act as a sensor of DNA replication. A second gene, *DPB11,* was isolated as a multicopy suppressor of *pol2* and *dpb2*, which encodes two essential subunits of DNA polymerase II. *DPB11* is essential for cell proliferation. Thermosensitive(*ts*)*dpb11-1* cells are defective in DNA replication and in the checkpoint response to replication blocks at the restrictive temperature. Physical interaction between Dpb11 and Pol2 suggests that Dpb11 is part of the DNA polymerase II complex and is required for the checkpoint response. The *S.pombe* homolog of Dpb11, Cut5, has also been shown to play a role in DNA replication and the S-phase checkpoint. Both Dpb11 and Cut5 have BRCT domains, which was first described in breast cancer protein BRCA1 and has been found in many checkpoint proteins such as Rad9, Crb2, etc. The function of the BRCT domain is unknown, but

some evidence suggest that the BRCT domain is involved in protein interaction or DNA binding.

To gain insight on the mechanism of the S-phase checkpoint pathway, and on Dpb11's involvement in this process, I carried out a genetic screen looking for high copy suppressors of *dpb11-1(ts)*. These suppressors might have a redundant function with Dpb11, or play a role downstream of Dpb11. *SDT1*(suppressor of *dpb11-1(ts)*) was identified in this screen. High copy *SDT1* not only suppress *dpb11-1(ts)*, but also weakly suppress the growth defect of *pol2* mutant at restrictive temperature, suggesting *SDT1* has genetic interaction with both *DPB11* and *POL2*. *SDT1* is essential for cell proliferation. Its mRNA level is cell-cycle regulated and peaks at the G1/S transition. Additionally, the *sdt1-1* mutant displays a defect in S-phase progression at 37°C. These observations indicate a requirement for Sdt1 during DNA replication. A role in the S-phase checkpoint is also suggested by other mutant alleles of *SDT1*. In the presence of a DNA replication block, these mutants enter anaphase prematurely, leading to lethality. These mutant phenotypes imply that Sdt1 functions closely with Dpb11. This is further supported by the fact that overexpression of DPB11 can also suppress the *ts* phenotype of *sdt1-1*.

My current goal is to further characterize the replication and checkpoint functions of Sdt1. Additionally, I am testing if there is a physical interaction between Sdt1 and Dpb11, whether the BRCT domain of Dpb11 is important for this interaction, and whether such interaction is required for the S-phase checkpoint signaling.

f. Siu Sylvia Lee, Graduate School of Biomedical Sciences, Ronald T. Javier, Ph.D., Department of Molecular Virology, "Protein interactions of the oncoprotein Ad9E4ORF1".

ABSTRACT

Human adenovirus type 9 (Ad 9) uniquely elicits estrogen-dependent mammary tumors when inoculated into rats. One of the primary oncogenic determinants is the transforming protein encoded by the Ad9 E4 region open reading frame 1 (9ORF1) gene. I have previously discovered that the C-terminus of 9ORF1 encodes a consensus PDZ domain-binding motif and 9ORF1, via this consensus motif, interacts with two PDZ domain-containing cellular factors: hDlg/SAP97 (DLG), a mammalian homolog of the Drosophila disc large tumor suppressor protein Dlg-A, and 9BP-1, a novel cellular factor.

I was in the process of cloning the full-length 9BP-1 cDNA, when its rat homology was cloned and published recently. Rat 9BP-1 encodes a 2055 amino acid protein with 13 consecutive PDZ domains and no other recognizable functional domains. I subsequently obtained the rat 9BP-1 cDNA and generated mammalian expression constructs for either N-terminal HA-tagged or untagged wild-type 9BP-1. In transiently transfection assays, I confirmed the *in vitro* and *in vivo* interactions between 9ORF1 and 9BP-1.

I have also generated a rabbit polyclonal 9BP-1 antiserum against the C-terminal 526 amino-acid residues of the protein. Using indirect immunofluoresence, I detected diffuse cytoplasmic and some cell-cell contact staining for 9BP-1 in normal CREF cells (established rat embryo fibroblast). In CREF cells expressing wild-type 9ORF1, 9BP-1 redistributed into cytoplasmic clusters (in >95% of cells), reminiscent of those observed for the

9ORF1 protein. In CREF cells expressing a 9ORF1 Region III mutant (mutIII A), which fails to associate with 9BP-1, 9BP-1 localization was not altered. Similar alteration of 9BP-1 subcellular localization was observed in C127 cells (~90%) stably expressing 9ORF1, as well as ~20% of TE85 and MCF7 expressing cells. In general, PDZ-domain proteins facilitate signal transduction by functioning as adaptors to assemble membrane receptors and cytosolic signalling molecules into multiprotein complexes at specialized regions of cell-cell contact. Our results suggest that 9ORF1 may disrupt the function of a select group of PDZ-domain proteins that regulate cellular proliferation by sequestering them within cytoplasmic aggregates in cells.

B. Enhancement Programs

Two education programs specific for this training program have been functional for the past four years. The biweekly journal club in which students provided literature reviews and faculty/postdoctoral fellows provided research reviews for 1997 - 1998 is shown in Table II.

The course in "Molecular Carcinogenesis" is given every Winter bloc and each student has successfully passed (pass requires a grade of B or better) this course. The topic outline for this course is shown in Table III.

C. Trainee Progress Review

Prospective trainees are nominated by letter and supporting documentation by a faculty member of the training program. The material on the prospective nominees are reviewed and the trainee selected by a standing subcommittee. Research progress of the trainees is reviewed by a standing departmental thesis committee, six faculty, which includes the major advisor and at least one other member of the training grant. This committee meets twice yearly and submits a written report to the graduate school.

7. CONCLUSIONS

The training program in breast cancer functioned as planned. Six students were enrolled in the training program at any given time. The breast cancer journal club met every two weeks and the involvement of additional laboratories increased every year due to new funding on breast cancer. The outside speakers over the past three years were a tremendous success as it allowed trainees not only to hear state of the art research seminars but to interact with the great speakers in a one-and-one-half-hour student only session with the speakers. The guest speakers, financially supported by the Department of Cell Biology resource, included Drs. Mina Bissell (University of California, Berkeley), Gilbert Smith (National Cancer Institute), Barbara Weber (University of Pennsylvania), Dennis Slamon (University of California, Los Angeles), Clive Dickson, Imperial Cancer Research Fund, London), Wen-Hwa Lee and Jolene J. Windle, (University of Texas Health Science Center, San Antonio), William Muller (McMaster University, Ontario), Satyabrata Nandi (University of California, Berkeley) and Malcolm Pike (University of Southern California). The publication productivity of the trainees is listed below and includes a) publications, b) meeting abstracts and c) manuscripts currently in final phases of preparation.

8. PUBLICATIONS/MEETING ABSTRACTS

Publications:

Said, T.K., Bonnette, S., and Medina, D. Immortal, non-tumorigenic mouse mammary outgrowths express high levels of cyclin B1 and activation of cyclin B1/cdc2 kinase. Cell Prolif. 29:623-639, 1996.

Stepanova, L., Leng, X. and Harper, J.W. Analysis of mammalian Cdc 37, a protein kinase targeting subunit of hsp 90. Methods Enzymol. 283:220-229, 1997.

Weiss, R.S., Gold, M.O., Vogel, H., and Javier, R.T. Mutant adenovirus type 9 E4 ORF1 genes define three protein regions required for transformation of CREF cells. J. Virol. 71:4385-4394, 1997.

Lee, S.S., Weiss, R.S., and Javier, R.T. Binding of human virus oncoproteins to hDlg/SAP97, a mammalian homolog of the Drosophila discs large tumor suppressor protein. Proc. Natl. Acad. Sci. USA 94:6670-6675, 1997.

Weiss, R.S. and Javier, R.T. A carboxy-terminal region required by the adenovirus type 9 E4 ORF1 oncoprotein for transformation mediates direct binding to cellular polypeptides. J. Virol. 71:7873-7880, 1997.

Jones, J.M., Attardi, L., Godley, L.A., Laucirica, R., Medina, D., Jacks, T., Varmus, H.E., Donehower, L.A. Absence of p53 in a mouse mammary tumor model promotes tumor cell proliferation without affecting apoptosis. Cell Growth & Differentiation 8:829-838, 1997.

Leng, X.-H., Connell-Crowley, L., Goodrich, D. and Harper, J.W. S-phase entry upon ectopic expression of G1-cyclin-dependent kinases in the absence of retinoblastoma protein phosphorylation. Current Biology 7:709-712, 1997.

Gangolli, E.A., Conneely, O.M. and O'Malley, B.W. Neurotransmitters activate the human estrogen receptor in a neuroblastoma cell line. J. Ster. Biochem. and Cell Biol. 61:1-9, 1997.

Bonnette, S., Kittrell, F.S., Stephens, L.C., Meyn, R.E. and Medina, D. Interactions of apoptosis, proliferation and host age in the regression of a mouse mammary preneoplasia (submitted for publication).

Abstracts

Roy, D., Contreras, A., Coleman-Krnacik, S. and Rosen, J.M. Isolation of molecular markers for ETBS in mammary gland development. 9th International Conference on Carcinogenesis and Risk Assessment. Etiology of Breast and Gynecological Cancers, Austin, Texas, 1995.

Chua, S.S., Wang, Y., DeMayo, F.J., and Tsai, S.Y. A bitransgenic mice model for breast cancer. Cancer Genetics and Tumor Suppressor Genes, Cold Spring Harbor, NY, August 14-18, 1996.

Gangolli, E.A., Conneely, O.M. and O'Malley, B.W. Neurotransmitters synergize to activate the human estrogen receptor in a neuroblastoma cell line. Tenth International Congress in Endocrinology, San Francisco, CA, P1:61, 1996.

Jones, J.M. and Donehower, L.A. The role of p53 loss in the acceleration of mammary tumor growth in the p53-deficient/wnt-1 transgenic mouse. Eighth Intl. p53 Workshop, Dundee, Scotland, 1996.

Bonnette, S.G., and Medina, D. Characterization of the TGF-β1 response in a mouse mammary epithelial cell line and its transformed counterpart. Gordon Conference on Mammary Gland Biology, Plymouth State College, Plymouth, NH, June 15-20, 1997.

Lindsey-Wyszomierski, S., Yeh, J. And Rosen, J.M. Glucocorticoid receptor/STAT5 interactions at the β-casein promoter. Keystone Meeting on Signal Transduction, February, 1998.

Lee, S.S. and Javier, R. The adenovirus type 9 E4ORF1 oncoprotein mislocalizes cellular PDZ-domain proteins. Cold Spring Harbor Symposium in "Pathways to Cancer". March, 1998.

Manuscripts in Preparation:

Wyszomierski, S.L., Yeh, J. and Rosen, J.M. STAT5/Glucocorticoid receptor protein: protein interactions modulate the activation state of STAT5 (in preparation).

Kazansky, A.V., Kobotyanski, E.B., Yeh, J., Wyszomierski, S.L. and Rosen, J.M. Differential activation of STAT5 isoforms by *src* family kinases and prolactin (in preparation).

Jones, J.M., Medina, D., Leder, P., Varmus, H.E., Donehower, L. Reduction in *p21* levels sufficient to accelerate mammary tumor growth rate in the *p21*-deficient/*Wnt-1* transgenic mice (in preparation).

Table I. Status of Trainees Previously Supported by DOD Training Grant

Student	Department	Advisor	Thesis Project	Status
Esha A. Gangolli	Cell Biology	B. W. O'Malley	Generation of a progesterone receptor transgenic mouse model	Graduated Fall, 1996. Current post-doctoral fellow; Univ. of Washington, Seattle.
Sharon G. Bonnette	Cell Biology	D. Medina	Mechanism of TGFβ1 inhibition of mammary cell growth	Graduated March, 1998; Current postdoctoral fellow
Jeffrey M. Jones	Molecular Virology	L. Donehower	Role of p53 in mammary tumorigenesis	Scheduled to graduate August, 1998.
Annette C. Hollman	Molecular Virology	J. Butel	Oncogene co-operativity in mammary tumorigenesis	Scheduled to graduate June, 1999.
Steven Chua	Cell Biology	M.-J. Tsai	Development of a bitransgenic mouse system to study oncogene function	Scheduled to graduate June, 1999.
Deana L. Roy	Cell Biology	J. Rosen	Molecular markers for terminal end buds in mammary gland development	Special status graduate student (see text).
Robin Weiss	Molecular Virology	R. Javier	Role of adenovirus type 9E4ORF1 in mammary oncogenesis	Graduated, Summer, 1997; Current postdoctoral fellow at Harvard.

Table II.

BREAST CANCER JOURNAL CLUB SCHEDULE
1997 - 1998

SEMINAR DATE	TIME	ROOM #	SPEAKER	DEPARTMENT
09/24/97	12:00 - 1:00 pm	M616	Michael Mixon	Cell Biology
10/08/97	12:00 - 1:00 pm	M616	Lakshmi Sivaraman	Cell Biology
10/29/97	12:00 - 1:00 pm	M616	Shannon Lindsey	Cell Biology
11/12/97	12:00 - 1:00 pm	M616	Sophia Tsai	Cell Biology
11/19/97	**4:00 - 5:00 pm**	**M112**	Jolene Windle, Ph.D.,Guest Lecturer	San Antonio
12/10/97	12:00 - 1:00 pm	M616	Dan Medina (Cancelled)	Cell Biology
01/07/98	12:00 - 1:00 pm	M616	Jeff Jones	Molecular Virology
01/21/98	12:00 - 1:00 pm	M616	Kristen Murphy	Cell Biology
02/04/98	12:00 - 1:00 pm	M616	Ron Javier	Molecular Virology
02/18/98	12:00 - 1:00 pm	M616	Sharon Plon	Pediatrics
03/04/98	**4:00 -5:00 pm**	**M112**	Wm. J. Muller, Ph.D.,Guest Lecturer	McMaster University
03/18/98	12:00 - 1:00 pm	M616	Xiao-Hong Leng	Biochemistry
03/25/98	**4:00 -5:00 pm**	**M112**	Satyabrata Nandi, Ph.D., Guest Lecturer	Univ. Calif., Berkeley
04/15/98	12:00 - 1:00 pm	M616	Sylvia Lee	Molecular Virology
04/29/98	12:00 - 1:00 pm	M616	Barry Markaverich	Cell Biology
05/13/98	**4:00 - 5:00 pm**	**M112**	Malcolm Pike, Ph.D., Guest Lecturer	USC Medical Center
05/20/98	12:00 - 1:00 pm	M616	Hong Wang	Biochemistry

Table III.

INTRODUCTION TO MOLECULAR CARCINOGENESIS

(L. Donehower, D. Medina)

This course explores the fundamental concepts and experiments in tumor biology, cancer virology and molecular oncogenesis. It provides a broad based extension of the introductory cancer material provided to students in the Complex Systems core curriculum course. It is designed for students who want a more in-depth treatment of the biology and molecular biology of cancer. Faculty from five departments serve as instructors. The course is open to graduate students and clinical fellows with a minimum requirement of Biochemistry or Cell Biology. In addition, the core curriculum course in Complex Systems is highly recommended, though not required. Lectures will be given on Mondays, Wednesdays, and Fridays at 1:00-2:00 p.m.

LECTURE	DATE	TOPIC	INSTRUCTOR	ROOM
1	3/16/98	Introduction/Epidemiology	DM	N317
2	3/18/98	Cell Cycle and Cancer	WH	N317
3	3/23/98	Growth Factors 1: Stimulatory	LY	N317
4	3/25/98	Growth Factors II: Inhibitory	LY	N311
5	3/27/98	Oncogenes I: Membrane Signalling	LD	N317
6	3/30/98	Oncogenes II: Signal Transduction	LD	N317
7	04/01/98	Oncogenes III: Transcriptional Regulation	LD	N311
8	04/03/98	Tumor Suppressors I	BS	N311
9	04/06/98	Tumor Suppressors II	BS	N317
10	04/08/98	Tumor Suppressors III	BS	N315
11	04/13/98	Apoptosis and Cancer	LD	N317
12	04/15/98	DNA Repair Genes and Cancer	HY	N317
13	04/17/98	Viruses and Cancer: Intro/Adenoviruses	RJ	N317
14	04/20/98	Viruses and Cancer: Papovaviruses	JB	N317
15	04/22/98	Viruses and Cancer: Herpesviruses	PL	N317
16	04/24/98	Viruses and Cancer: Retroviruses	LD	N317
17	04/27/98	Tumor Progression and Metastases	DM	N317
18	04/29/98	Angiogenesis	DM	N317
19	05/01/98	Chemical Carcinogenesis	DM	N317
20	05/04/98	Animal Models and Cancer	LD	N317
21	05/06/98	Gene Therapy for Cancer	EA	N315
22	05/08/98	Reversion and Inhibition of Cancer	DM	N317
	05/13/98	FINAL (1-4 p.m.)	DM	N317

Lecturers

Daniel Medina * (DM)
Wade Harper * (WH)
Janet Butel * (JB)
Estuardo Aguilar-Cordova (EA)
Larry Donehower * (LD)
Hagop Youssoufian (HY)
Lynn Yeoman (LY)
Betty Slagle (BS)
Ron Javier * (RJ)
Paul Ling (PL)

* Indicates participating faculty on Institutional Training Grant in Breast Cancer.